

COLLEGE of AMERICAN PATHOLOGISTS

HER2 Testing and Clinical Decision Making in Gastroesophageal Adenocarcinoma

Guideline update from the College of American Pathologists, in collaboration with the American Society for Clinical Pathology

Teaching Presentation

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Archives of Pathology & Laboratory Medicine

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1 Background and Methodology

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Introduction

- In 2016, the American Society for Clinical Oncology (ASCO), the American Society for Clinical Pathology (ASCP), and the College of American Pathologists (CAP) published an evidence-based guideline on HER2 testing in gastroesophageal adenocarcinoma (GEA).
- Since then, both pathology testing practices and treatment strategies have continued to evolve.
- In 2024, the CAP convened an expert panel to review the literature, define key questions, and apply a standard methodological framework to update recommendations.

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Objective

- To evaluate the current literature and update evidence-based recommendations for HER2 assessment in GEA to reflect evolving specimen types, treatment pathways, and testing technologies.

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Target Population and Audience



Target Population

Patients with advanced GEA

- Treatment naive
- Those being considered for subsequent therapy after disease progression



Target Audience

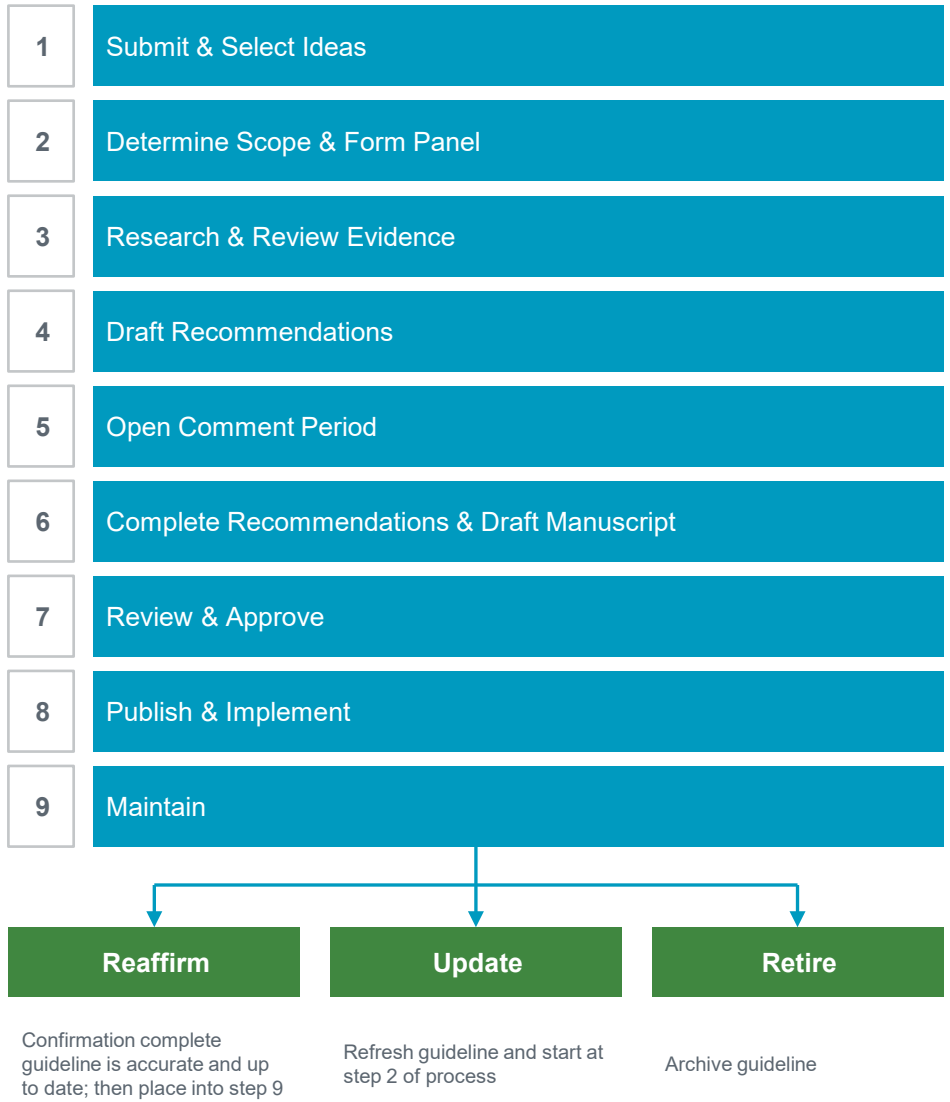
Pathologists, gastroenterologists, oncologists, histotechnologists, hospital or laboratory managers, and patient advocacy group representatives

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Guideline Development Process

- Multi-Society Collaboration
  - CAP, ASCP, and ASCO
- Transparency
  - Aligned with National Academy of Medicine-endorsed standards
  - PROSPERO-registered protocol
- Consistency
  - Recommendations developed using the Grading of Recommendations Assessment, Development, and Evaluation (GRADE) approach

Evidence-Based Guideline Development Methodology Manual



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Key Questions (KQs)

KQ1 In specimens from patients with advanced GEA, do HER2 test results from other sites or specimens (resection specimens, tissues from metastatic sites, FNA/cytology specimens from primary or metastatic tumors, liquid biopsies) yield concordant results when compared to primary biopsy specimens?

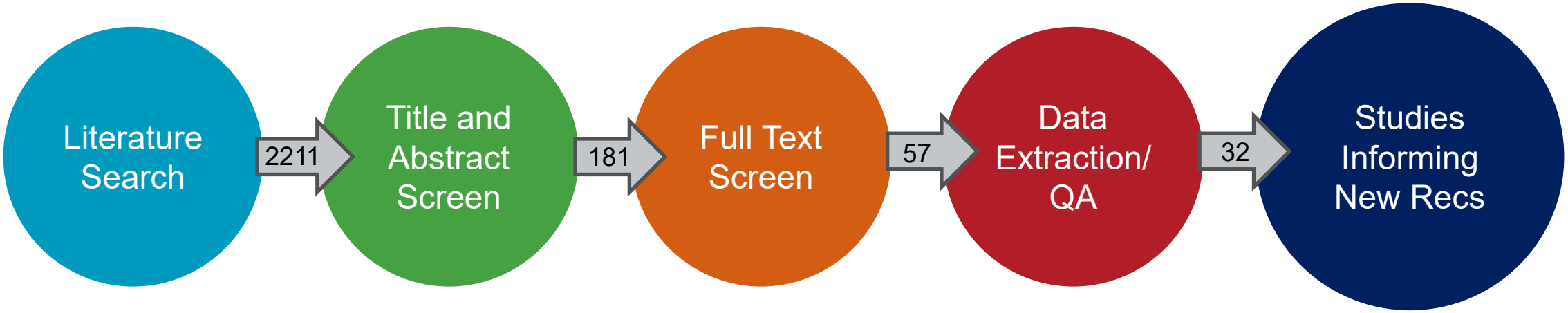
KQ2 In specimens from patients with advanced GEA, how do the test characteristics of genomic testing (NGS/PCR across tissue-based and blood-based platforms) compare with traditional HER2 assessment methodologies (IHC with different antibodies, ISH with various probe platforms)?

KQ3 In patients with advanced HER2-positive GEA being considered for subsequent therapy after disease progression, does repeat HER2 testing inform further treatment decisions compared to not repeating testing?

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Literature Search & Systematic Review

- Conducted in Ovid MEDLINE, Embase, Cochrane Library
- Search period: January 1, 2015, to November 6, 20242



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Results



The expert panel conducted a systematic review assessing 2,211 titles to address the key questions.



Three new strong recommendations were made



Four statements from the 2016 guideline were reaffirmed as good practice statements

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2 Recommendations and Rationale

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Statement 1 (KQ1: Which specimen?)

Strong Recommendation

At initial diagnosis of advanced GEA, treating clinicians or pathologists should request HER2 testing on the highest quality tumor specimen (primary or metastasis). Pathologists should select the tissue block with the areas of lowest grade tumor morphology in biopsy, resection, or FNA specimens. More than one tissue block may be selected if different morphologic patterns are present.

Note: Highest quality tumor for HER2 testing in GEA = high cellularity, minimal necrosis, large viable area, low grade, no treatment effect.

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Specimen Type and Tumor Heterogeneity

- HER2 testing in GEA is influenced by specimen type and tumor heterogeneity.
- Biopsy–resection concordance is generally >90%, though false negatives can occur.
- Tumor content within biopsy fragments is more important than the number of fragments.

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Block Selection and Metastatic Specimens

- Select blocks with high tumor cellularity, minimal necrosis, and no treatment effect.
- Testing more than one block increases HER2 detection in heterogeneous tumors.
- Primary–metastatic concordance is usually >80%; HER2 status conversion can occur.
- Metastatic tissue may be tested when quality is comparable or when clinically relevant.

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Cytology and Preanalytical Considerations

- Cytology specimens may be used when tissue is unavailable, but interpretation is challenging.
- Performing ISH in malignant effusions may improve reliability.
- Fixation and cold ischemic time affect HER2 results; >6 hours cold ischemia degrades IHC/ISH.
- HER2 testing should be performed on invasive carcinoma, not dysplasia.

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Statement 2 (KQ2: Which test?)

Strong Recommendation

In specimens from patients with advanced GEA, pathologists should use IHC with subsequent ISH as needed for the primary assessment of HER2 status. Genomic testing (liquid and/or solid) may be used concurrently or subsequently for clinical decision making.

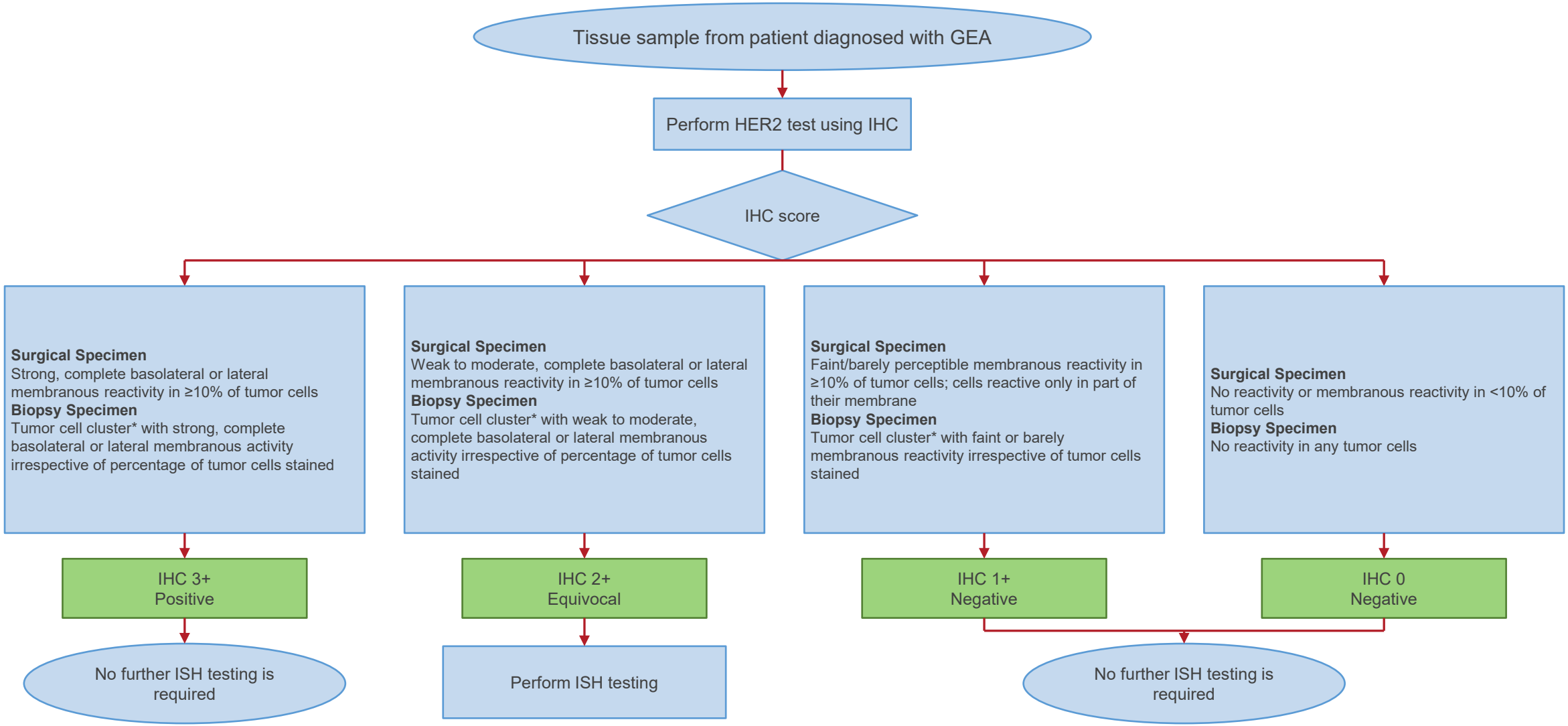
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HER2 Testing Algorithm (Unchanged From 2016)

- HER2 IHC should be performed first, with reflex ISH for equivocal (2+) cases.
- IHC 3+ (positive) and IHC 0/1+ (negative) results do not require ISH.
- Rüschoff-Hoffmann scoring criteria should be used for HER2 IHC and ISH.
- This IHC-centric approach remains the standard for determining HER2 status in advanced GEA.

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Algorithm



* Tumor cell cluster: a cohesive cluster of 5 or more neoplastic cells showing membranous HER2 reactivity.

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Why IHC/ISH Remains Preferred

- Tissue-based IHC/ISH is more sensitive and reliable than alternative genomic testing for HER2 in GEA.
- Reported concordance between NGS and IHC/ISH is variable, with lower agreement among HER2-positive cases.
- Multiple studies show false-negative HER2 results by NGS compared with IHC/ISH.
- Tumor heterogeneity and low-level amplification contribute to reduced NGS sensitivity.

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IHC/ISH vs Genomic Testing: HER2 Concordance

Wide range of reported concordance

- Reported concordance between NGS-based HER2 assessment and IHC/ISH ranges widely across published studies.
- Example: Janjigian et al 2020 vs Ross et al 2017 reported overall concordance of 68% and 100%, respectively (same NGS platform/institution).

Tissue-based genomic testing has low sensitivity

- Reported sensitivity of tissue-based genomic testing compared to IHC/ISH ranged from 50%–70%, indicating potential false negatives relative to conventional HER2 assessment.

ct-DNA has even lower sensitivity

- Reported sensitivity of liquid biopsy (ctDNA) compared to IHC/ISH ranged from 20%–73%, suggesting an even higher risk of missing HER2 positivity.

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Role of Genomic Testing

- Genomic testing should not replace IHC/ISH for establishing HER2 status.
- NGS may be used in addition to IHC/ISH to support broader clinical decision making.
- IHC/ISH testing is more standardized and widely available, supporting diagnostic equity.
  - This approach is acceptable, feasible, and consistent with prior recommendations.

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Statement 3 (KQ3: Repeat testing?)

Strong Recommendation

In patients with advanced HER2-positive GEA being considered for subsequent therapy after disease progression, HER2 assessment may be performed on relapsed/recurrent tumor sample. Tissue is preferred but if not available/feasible, liquid testing may be performed.

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Why Reassess HER2 at Disease Progression?

- Prognosis after progression is poor, with limited second-line therapy options.
- Most trials of HER2-targeted agents failed to improve overall survival (OS) or progression-free survival (PFS).
- Recent phase 2 DESTINY-Gastric trials demonstrated benefit with trastuzumab deruxtecan in patients with persistent HER2 positivity.
- HER2 status after first-line therapy affects second-line treatment selection.

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Loss of HER2 Positivity After Trastuzumab Therapy

Pietrantonio et al (2016)

- HER2 positivity loss in 32% (13/41 cases)

Saeki et al (2018)

- HER2 positivity loss in 60% (20/33 cases)

Seo et al (2019)

- HER2 positivity loss in 29% (14/48 cases)

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Loss of HER2 Positivity After Trastuzumab Therapy, continued

- Loss is more common in initially IHC 2+/ISH amplified tumors than IHC 3+ tumors.
- HER2 loss is associated with tumor heterogeneity, low initial H-scores, and genetic variability.
- HER2 status changes were defined using IHC and/or ISH on post-treatment tissue.
- HER2 loss is a recognized mechanism of acquired trastuzumab resistance.

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Clinical Impact of HER2 Loss

- Only one study (Seo et al) evaluated second-line therapy after HER2 loss.
  - Patients received trastuzumab emtansine (T-DM1):
    - 10 with persistent HER2 positivity
    - Three with loss of HER2 positivity
  - No statistically significant difference in ORR or PFS between the two cohorts.
  - Kaplan-Meier curve showed a "distinct tendency for worse prognosis in the loss of HER2-positivity group."
  - Statistical analysis limited by small sample size.

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3 Summary of Good Practice Statements (GPSs)

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2016 Guideline Statements Reaffirmed

- Four statements from the 2016 guideline were reaffirmed as GPSs.
- These statements did not undergo a new systematic review.
- Public comment supported reaffirmation (~80% agreement).
- The reaffirmed statements reflect foundational laboratory and pathology practices that remain current.

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Clarifications to Reaffirmed GPSs

- HER2 IHC scoring:
  - Continue using the Rüschoff-Hoffmann method.
  - Additional guidance (eg, microscope magnification strategies) may aid scoring accuracy.
  - Readers may refer to Scheel et al and prior studies (including ToGA) for further detail.
- HER2 ISH interpretation:
  - Apply standard ISH interpretation criteria when indicated.
  - HER2 amplification defined as HER2/CEP17 ratio ≥ 2.0, or
  - Average HER2 copy number ≥ 6 signals per cell (clarified wording).

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Statements Updated as GPS

2016 Guideline Statement	2025 Status
Laboratories/pathologists must specify the antibodies and probes used for the test and ensure that assays are appropriately validated for HER2 IHC and ISH on GEA specimens.	Reaffirmed without substantive change
When GEA HER2 status is being evaluated, laboratories/pathologists should perform/order IHC testing first, followed by ISH confirmation when IHC result is 2+ (equivocal). Positive (3+) or negative (0 or 1+) HER2 IHC results do not require further ISH testing.	Reaffirmed without substantive change

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Statements Updated as GPS, continued

2016 Guideline Statement	2025 Status
Pathologists should use the Rüschoff-Hoffmann method in scoring HER2 IHC and ISH results for GEA.	**Wording updated:** Pathologists should use the Rüschoff-Hoffmann method for HER2 IHC scoring in GEA and apply standard ISH interpretation criteria when indicated.
Laboratories must incorporate GEA HER2 testing methods into their overall laboratory quality improvement program, establishing appropriate quality improvement monitors as needed to ensure consistent performance in all steps of the testing and reporting process. In particular, laboratories performing GEA HER2 testing must participate in a formal proficiency testing program, if available, or an alternative proficiency assurance activity.	**Wording updated:** Laboratories should incorporate GEA HER2 testing methods into their overall laboratory quality improvement program, following the requirements of applicable local regulatory bodies and accreditation organizations.

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Scope Decisions for 2016 Statements

- Public comments opposing reaffirmation primarily concerned the two statements directed at treating clinicians.
- Because this revision focuses on pathologic recommendations, treatment recommendations from 2016 were archived.
- This update does not expand clinical management guidance beyond pathology scope.

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Statements Archived in Update

2016 Guideline Statement	2025 Status
In patients with GEA who are potential candidates for HER2-targeted therapy, the treating clinician should request HER2 testing on tumor tissue.	Archived[a]
Treating clinicians should offer combination chemotherapy and HER2-targeted therapy as the initial treatment for appropriate patients with HER2-positive tumors who have metastatic or recurrent GEA.	Archived[a]

[a] Archived because ASCO is a collaborator rather than a partnering organization for this guideline, and clinician-directed recommendations are covered in existing ASCO and National Comprehensive Cancer Network (NCCN) guidelines.

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4 Additional Information

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Additional Resources

- More information—including a supplement and clinical tools and resources—is available on cap.org.
  - HER2 Testing and Clinical Decision Making in Gastroesophageal Adenocarcinoma

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Update Expert Panel Members

Name	Expertise/Affiliation/Institution
Angela N. Bartley, MD, FCAP—Chair	Pathology/CAP/Banner Gateway, MD Anderson Cancer Center
Jaffer Ajani, MD	Oncology/ASCO/ University of Texas, MD Anderson Cancer Center
Julia A. Bridge, MD, FCAP	Pathology/CAP/University of Utah Health/ARUP
Amy Brownlee, MD, FCAP	Pathology/ASCP/ University of North Carolina, Chapel Hill
Kelsey E. McHugh, MD, FCAP	Pathology/ASCP/ Mayo Clinic-Arizona
Catherine Streutker, MD	Pathology/CAP/St. Michael's Hospital: Unity Health, Toronto
Zev Wainberg, MD	Oncology/ASCO/ UCLA Santa Monica Cancer Care
Mary Kay Washington, MD, PhD, FCAP	Pathology/CAP/Vanderbilt University Medical Center

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Resources

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